Standard Operating Procedure for Phytoplankton Sample Collection and Preservation Field Procedures

LG400

Revision 09, March 2002

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1.0 SCOPE AND APPLICATION

1.1 This standard operating procedure describes the sampling and preservation of phytoplankton samples taken for the GLNPO open water Great Lakes surveys.

2.0 SUMMARY OF METHOD

2.1 Integrated (INT) phytoplankton samples are created from a composite of water samples taken at discrete depths with the rosette from the euphotic zone of the water column. For an unstratified water column, the integrated sample is prepared by taking equal volumes of water from SRF (1 m), 5 m, 10 m and 20 meters unless the depth is less than 20 meters. If the total depth is between 15 and 22 meters, the 20 meter sample is replaced by the bottom sample (B-1 or B-2). If the total depth is less than 15 meters, equal volumes are taken from surface, mid-depth, and bottom sample (B-1 or B-2).

For a stratified water column, equal volumes are taken from the surface, 5 m, 10 m, and lower epilimnion (LEP). If the epilimnion is very shallow, equal volumes are taken from a maximum of four sampling depths and a minimum of two sampling depths. The underlying strategy is to collect a representative sample from the epilimnion.

2.2 Aliquots from each depth are combined, and approximately 1 L of the composite sample is preserved with Lugol's Solution for later analysis. Additional discrete samples from the deep chlorophyll layer (DCL) are taken during the summer at stations exhibiting marked horizontal discontinuities in chlorophyll concentration.

3.0 SAFETY AND WASTE HANDLING

3.1 Refer to GLNPO's *Health, Safety and Environmental Compliance Manual* (May 1997, or as amended) and individual instrument procedural operations manuals for specific details on applicable 1) personal health and safety issues; 2) instrumental, chemical, and waste handling procedures; and 3) accident prevention. This applies to all EPA personnel, EPA contractors or federal, state, or local government agencies, and persons who operate or are passengers onboard US EPA GLNPO vessels during all activities and surveys.

3.2 It is the responsibility of the user of this method to comply with relevant chemical disposal and waste regulations as sited in GLNPO's *Health, Safety and Environmental Compliance Manual* (May 1997, or as amended). All applicable safety and waste handling rules are to be followed. All containers storing reagents, standards, controls, blanks, and wastes used in the laboratory must be properly identified through appropriate labeling and hazard definition. Good technique includes minimizing contaminated waste. Over-board discharges of chemical wastes are forbidden.

3.3 Every chemical should be regarded as a potential health hazard and exposure to these compounds should be as low as reasonably achievable. Please refer to Appendix L in GLNPO's *Health,*

Safety and Environmental Compliance Manual (May 1997, or as amended) for more detailed descriptions of the potential risks associated with any chemicals used in this method. It is good laboratory practice to wear a lab coat, safety goggles or glasses and gloves at all times.

3.4 During sampling, caution, common sense and good judgement should dictate appropriate safety gear to be worn in any given situation on deck. Hardhats, gloves, and steel-toed shoes must be worn in working conditions where there is a possibility of injury to the head, hands, or feet; however, if in doubt, please ask the Chemical Hygiene Officer.

3.5 Collecting samples in cold weather, especially around cold water bodies, carries the risk of hypothermia and frostbite. Sampling team members should wear adequate clothing for protection in cold weather. For specific information regarding sampling during cold conditions, please refer to the US EPA GLNPO *Standard Operating Procedures for Winter Operations* (December 1994, or as amended).

3.6 Collecting samples in extremely hot and humid weather carries the risk of dehydration and heat stroke. Sampling team members should carry an adequate supply of water or other liquids for protection against dehydration in hot weather.

3.7 Work vests must be worn while working on the fantail and Rosette deck.

3.8 Preservation of the phytoplankton samples with Lugol's solution must take place in a hood, and gloves and safety glasses should be worn.

4.0 EQUIPMENT AND SUPPLIES

4.1 Equipment and supplies for sample collection and preservation

1000-mL plastic sample bottles
1-gallon cubitainers
Repipetter with 10-mL delivery capability
Lugol's solution

4.2 Equipment and supplies for preparation of Lugol's solution

Reagent water
Glacial acetic acid
Iodine
Potassium iodide (KI)
1-L Volumetric flask
Opaque 1-L container
Glass funnel

5.0 REAGENTS

5.1 **Lugol's Solution:** Prepared at least one week prior to survey

5.1.1 In a fume hood, dissolve, 100 g of KI and 50 g of I_2 in approximately 800 mL of reagent water in a 1-L volumetric flask. Mix until the chemicals are completely dissolved. Add 100 mL of glacial acetic acid and bring volume up to 1 L with reagent water.

5.1.2 This preservative is stored in an opaque bottle labeled with the contents, date of preparation, and preparer's initials.

6.0 SAMPLE COLLECTION AND PRESERVATION

NOTE: Step 6.1 is generally done by the ship contractor or EPA personnel. Other personnel working on related cooperative agreements or contracts will conduct this task when requested.

6.1 A 1-L composite sample is collected according to Rosette Sampler Procedure (LG 200 - section 4.2) at each station. In Lake Superior, two 1-L samples must be taken for each station due to the low numbers of organisms in the lake. During the summer survey, a DCL sample may be also be taken at stations exhibiting marked horizontal discontinuities in chlorophyll concentration. Depths at which the DCL samples are taken are determined by *in vivo* fluorescence profiles, as decided by EPA personnel.

6.2 In the Biology lab, add 10 mL of Lugol's solution to each 1-L water sample within 2 hours of sample collection to obtain a final concentration of 1%.

6.3 The neck of the bottle is wrapped several times with parafilm to preclude spillage and evaporation.

6.4 Information on depth of sample collection and time of preservation are recorded on the phytoplankton collection data sheet.

6.5 Samples are stored in the dark and under refrigeration.